RESULTS

**Quantifying leaf proteins at the continental scale**

*Study area:*

Wide coverage, varying almost independently in temp and precip (Fig 1a and b)

*Protein composition of the average eucalypt leaf.*

On average 64% associated with photosynthesis – 36% with Calvin Cycle, 22% with light reactions (Fig 2a)

This is a higher degree of dominance by top few proteins than observed in [comparison]. It obviously reflects that phot is the primary function of leaves plus that phot reactions require large amounts of particular proteins, rubisco and photosystems being the top-ranked

MS hardware as it was tuned detected ~2000 individual proteins total (i.e. down to within 99.X% of total protein) among which top 500 ranked captured 90% of total protein mass (Fig 2c)

**3.) Linking leaf protein abundances with environment and functional traits.**

Because individual proteins can be quantified, pairwise correlations can be calculated between them, and between any given protein and a range of environment-at-site and physiological quantities (Fig 2a).

Total protein amounts are strongly driven by temp and to a lesser extent rainfall (Fig 2a, d(i). Individual protein groups are all correlated positively with total protein to varying extent, implicating: a.) a general thermodynamic requirement for greater amounts per leaf area of all major protein functional classes at lower temperatures, and b.) substitution of water use efficiency for N-use efficiency at low rainfall.

Per leaf area trends in CC’s are essentially identical to environmental trends in leaf protein abundance (cor = 0.97). We see strong declines in CC’s per leaf area in response to MAT (2b-i), and to a lesser extent MAP (2b-ii). Photosystems are less tightly correlated to total protein abundance (0.82) but we still see the MAT response (2b-i). Any potential MAP response is offset by a shift in the proportional abundance (i.e. fraction of total leaf protein) of photosystem proteins. CC’s do not respond to irradiance on a per leaf area basis, while a pronounced decline (how many %?) in photosystem protein abundance is evident in response to increasing light availability. This decline in photosystem protein is also apparent on a proportional basis; Calvin cycle protein abundance also increases slightly in response to irradiance (%).

The degree of intraspecific variation in photosystem protein proportional abundance is considerably higher than for Calvin cycle proteins. Leaves appear to be able to alter protein allocation to photosystems in response to environmental conditions (some stats and numbers). Calvin cycle proteins are somewhat more abundant than photosystem proteins (mean 33% vs 21%), which may account for some of this difference, but clearly not all of it.

Adjustments in per leaf area CC abundances are to some extent occurring via LMA: CC’s per leaf area increase with increasing LMA, although there is substantial variation in the CC – LMA relationship, indicating that LMA is responding to other requirements than photosynthetic capacity (see last para). Photosystem abundance does not increase on a per leaf area basis as leaves become thicker/denser, and reduces as a proportion of total leaf protein.

Something about leaf N, tighter correlations with Calvin cycle proteins than photosystems…

How does this section fit in?

The role of LMA vs protein concentration (i.e. as a fraction of leaf dry mass) in determining per leaf area protein abundance depends interactively on MAP and MAT. Low per leaf area protein abundance at warm, wet sites is more closely associated with low LMA than low protein concentration, while high per leaf area protein abundance at cool, dry sites is strongly associated with high protein concentration. (This isn’t anything that couldn’t have been done using LMA, leaf N% and leaf N\_area, but the point to make is that it’s not all just about increasing carboxylation capacity by adding layers of mesophyll).

1. Absolute amounts of protein per leaf area adjust along physical gradients as follows (lower row of Fig 2a):
   1. Light reactions decline at higher irradiance but calvin cycle doesn’t change (in other words, at lower irradiance there is more light-capture apparatus relative to CC)
   2. All types decline toward higher temp
   3. toward lower rainfall CC increases, no change to light reactions
   4. each of these make sense for reasonably-well-understood reasons
   5. combined effects in lower row of Fig 3d

No strong effect of environment on proportional allocation of CC’s (although some response to irradiance). Some evidence that carboxylation capacity per leaf area is increased by increasing LMA, although there is substantial variation in the total protein – LMA relationship, indicating that LMA is responding to other requirements than photosynthetic capacity (see last para).